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- (88) Date of publication of the international search report:  
21 October 2004
- For two-letter codes and other abbreviations, refer to the "Guidance Notes on Codes and Abbreviations" appearing at the beginning of each regular issue of the PCT Gazette.*

(54) Title: METHOD TO MEASURE A T CELL RESPONSE AND ITS USES TO QUALIFY ANTIGEN-PRESENTING CELLS

(57) Abstract: A method to characterize a T-cell response of a final population of T-lymphocytes resulting from the co-incubation of an initial population of T lymphocytes with a composition of antigen-presenting cells (APCs), said method comprising the steps of a) simultaneous measuring on a single cell basis at least two parameters: (i) proliferation of T lymphocytes and (ii) presence of a T cell antigen receptor on the surface of T lymphocytes and/or presence of at least one biological molecule produced by T lymphocytes, and the attribution of a positive or a negative value to each of these parameters, and b) classification of the final T-lymphocytes population into 2<sup>n</sup> different subsets of T lymphocytes, n being the number of parameters, each subset being characterized by a positive or a negative value respectively to each parameter, and the determination of the proportion of T lymphocytes present in each subset with respect to the number of T lymphocytes in the final population, with said proportion being characteristic of the T-cell response. Use of the method for batch release assay and potency assay.

WO 2004/050909 A3

# INTERNATIONAL SEARCH REPORT

International Application No  
EP 03/13579

**A. CLASSIFICATION OF SUBJECT MATTER**  
IPC 7 G01N33/50

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)  
IPC 7 G01N

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the International search (name of data base and, where practical, search terms used)

EPO-Internal, BIOSIS

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	GIVAN ALICE ET AL: "A flow cytometric method to estimate the precursor frequencies of cells proliferating in response to specific antigens" CYTOMETRY SUPPLEMENT, no. 10, 2000, page 94, XP002278599 & THE XX CONGRESS OF THE INTERNATIONAL SOCIETY FOR ANALYTICAL CYTOLOGY; MONTPELLIER, FRANCE; MAY 20-25, 2000 ISSN: 1046-7386 the whole document ----- -/--	1-44, 46-49

☒ Further documents are listed in the continuation of box C.

☐ Patent family members are listed in annex.

### \* Special categories of cited documents :

- \*A\* document defining the general state of the art which is not considered to be of particular relevance
- \*E\* earlier document but published on or after the International filing date
- \*L\* document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- \*O\* document referring to an oral disclosure, use, exhibition or other means
- \*P\* document published prior to the International filing date but later than the priority date claimed

\*T\* later document published after the International filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

\*X\* document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone

\*Y\* document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

\*G\* document member of the same patent family

Date of the actual completion of the International search

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# INTERNATIONAL SEARCH REPORT

International Application No

P 03/13579

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	<p>ALLSOPP C E M ET AL: "A flow cytometric method to assess antigen-specific proliferative responses of different subpopulations of fresh and cryopreserved human peripheral blood mononuclear cells" JOURNAL OF IMMUNOLOGICAL METHODS, ELSEVIER SCIENCE PUBLISHERS B.V.,AMSTERDAM, NL, vol. 214, no. 1-2, 1 May 1998 (1998-05-01), pages 175-186, XP004129949 ISSN: 0022-1759 abstract page 178, left-hand column, paragraph 2 - page 181, right-hand column, paragraph 1; figures 2-4</p>	1-44, 46-49
X	<p>----- DATABASE BIOSIS 'Online! BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US; 16 November 2002 (2002-11-16), ZAKI MOHAMED H ET AL: "Non-Neoplastic T Cells from B-CLL Patients Exhibit Intrinsic Defects in TCR Signaling." XP002278601 Database accession no. PREV200300336083 abstract &amp; BLOOD, vol. 100, no. 11, 16 November 2002 (2002-11-16), page Abstract No. 1483, 44TH ANNUAL MEETING OF THE AMERICAN SOCIETY OF HEMATOLOGY; PHILADELPHIA, PA, USA; DECEMBER 06-10, 2002 ISSN: 0006-4971</p>	1-44, 46-49
X	<p>----- DATABASE BIOSIS 'Online! BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US; 16 November 2002 (2002-11-16), RUBIO MARIE T ET AL: "Use of CFSE and Intracellular Cytokine Staining To Detect CD8 T Cell Response to Exogenous Protein Antigen Presented by Monocyte-Derived Human Dendritic Cells: Differential Effects of PGE2 on Antigen Presentation to CD4 Versus CD8 T Cells." XP002278602 Database accession no. PREV200300356793 abstract -/--</p>	1-44, 46-49

## INTERNATIONAL SEARCH REPORT

International Application No.

PCT/EP 03/13579

## C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
P,X	<p>&amp; BLOOD, vol. 100, no. 11, 16 November 2002 (2002-11-16), page Abstract No. 2659, 44TH ANNUAL MEETING OF THE AMERICAN SOCIETY OF HEMATOLOGY; PHILADELPHIA, PA, USA; DECEMBER 06-10, 2002 ISSN: 0006-4971</p> <p>-----</p> <p>BERCOVICI N ET AL: "Multiparameter precursor analysis of T-cell responses to antigen" JOURNAL OF IMMUNOLOGICAL METHODS, ELSEVIER SCIENCE PUBLISHERS B.V.,AMSTERDAM, NL, vol. 276, no. 1-2, 1 May 2003 (2003-05-01), pages 5-17, XP004422637 ISSN: 0022-1759 the whole document</p> <p>-----</p>	<p>1-44, 46-49</p>
A	<p>BARREDA DANIEL R ET AL: "Flow cytometric analysis of PKH26-labeled goldfish kidney-derived macrophages" DEVELOPMENTAL AND COMPARATIVE IMMUNOLOGY, vol. 24, no. 4, June 2000 (2000-06), pages 395-406, XP002278600 &amp; ISSN: 0145-305X abstract page 398, left-hand column, paragraph 2 - page 399, left-hand column, paragraph 1; figures 2-4</p> <p>-----</p>	<p>1-44, 46-49</p>

# INTERNATIONAL SEARCH REPORT

International application No.  
PCT/EP 03/13579

## Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. ☒ Claims Nos.:  
because they relate to subject matter not required to be searched by this Authority, namely:  
see FURTHER INFORMATION sheet PCT/ISA/210
2. ☒ Claims Nos.: 45  
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:  
see FURTHER INFORMATION sheet PCT/ISA/210
3. ☐ Claims Nos.:  
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

## Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this International application, as follows:

1. ☐ As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.
2. ☐ As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.
3. ☐ As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:
4. ☐ No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

### Remark on Protest

- ☐ The additional search fees were accompanied by the applicant's protest.
- ☐ No protest accompanied the payment of additional search fees.

## FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

## Continuation of Box I.1

## Rule 39.1(v) PCT - Presentation of information

Claims 1 to 26 concern a method of characterizing a T-cell response of a final population of T-lymphocytes which method defines the parameters to be measured and how data is to be treated. The result of the method is the characterization of the final population of T-lymphocytes by classification into a number of subsets related to the number of parameters measured. Said method however lacks a technical character because the subject-matter of each claim as a whole does not cause any technical effect.

The search has been directed to uses of the methods as defined in claims 27 to 44 and 46 to 49.

## Continuation of Box I.2

Claims Nos.: 45

No text is present under the heading "Claim 45". Thus, no search was carried out for claim 45.

The applicant's attention is drawn to the fact that claims relating to inventions in respect of which no international search report has been established need not be the subject of an international preliminary examination (Rule 66.1(e) PCT). The applicant is advised that the EPO policy when acting as an International Preliminary Examining Authority is normally not to carry out a preliminary examination on matter which has not been searched. This is the case irrespective of whether or not the claims are amended following receipt of the search report or during any Chapter II procedure. If the application proceeds into the regional phase before the EPO, the applicant is reminded that a search may be carried out during examination before the EPO (see EPO Guideline C-VI, 8.5), should the problems which led to the Article 17(2) declaration be overcome.